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Occurrence and distribution of Rutaceae in protected areas of Espírito Santo state, Brazil: a survey integrating specimen occurrence and remote sensing data

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Abstract

The state of Espírito Santo, Southeastern Brazil, is renowned for its species biodiversity, especially because of the Atlantic Forest, a globally recognized hotspot. Despite this significance, the conservation status of many species remains uncertain due to the state's limited protected areas network (PAs) and incomplete species knowledge within Espírito Santo's PAs. In this study, we worked on a comprehensive dataset of preserved specimen occurrences of Rutaceae within Espírito Santo, integrating remote sensing data, preserved specimen occurrences and PA delimitations. Satellite images from Landsat-8 were obtained and analyzed, focusing on bands representing distinct segments of the electromagnetic spectrum. The Normalized Difference Vegetation Index (NDVI) was obtained to assess vegetation coverage and land use profiles. We found that a total of 62 species native to Brazil in 17 genera of Rutaceae have records within Espírito Santo state. The majority of occurrence records in our dataset were located outside Espírito Santo PAs (1,634 records, accounting for 68% of the total 2,386 reported and georeferenced occurrences), and there are 21 species not represented within any of the known PAs of Espírito Santo. Among these unrepresented species, six are endemic to the state and/or are threatened with extinction, all belonging to the genus *Conchocarpus*. Our discussion highlights the need for expanded field expeditions, improved funding, and strategic sampling within PAs to better understand and protect Espírito Santo's and the Atlantic Forest biodiversity. Additionally, the use of remote sensing data provides insights into vegetation health and can guide future conservation efforts.

 $\textbf{Keywords} \ \ \text{Biogeography} \cdot \text{Atlantic Forest} \cdot \text{Online databases} \cdot \text{Phytogeographic domains} \cdot \text{Vegetation types}$

1 Introduction

Brazil's tropical forests, which include the vast Amazon rainforest and the predominantly confined Atlantic Forest, rank among the world's richest in terms of species richness and endemism (Baker et al. 2020; Peres et al. 2020; Marques et al. 2021; Rezende et al. 2021). Historical connections have

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been documented between these once-continuous forested biomes (Ledo and Colli 2017; Fine and Lohmann 2018). The Atlantic Forest, recognized as a global biodiversity hotspot (Mittermeier et al. 1999; Myers et al. 2000), is one of Earth's most biologically rich yet critically endangered regions. Furthermore, the southeastern portion of the Atlantic Forest is considered a "darkspot," i.e., an area predicted to harbor numerous undescribed and unrecorded species (Ondo et al. 2024). This threatened status in Brazil stems from several factors, including habitat fragmentation and deforestation, both rooted in Brazil's colonial history and exacerbated by the region's high population density and extensive urbanization (Ribeiro et al. 2011; Tabarelli et al. 2012; Rezende et al. 2018). Additionally, while biodiversity data have become increasingly accessible online, the documentation of Atlantic Forest's biodiversity is still challenging, particularly in the availability of occurrence records in biodiversity datasets (Colli-Silva et al. 2020).



To safeguard Atlantic Forest's unique biodiversity in Brazil, strategies to establish a protected areas network (henceforth PAs) within the country have been formulated, aiming to bolster sustainable land usage and biodiversity conservation. Many strategies have yielded positive outcomes, with current data indicating approximately 28% of remaining native vegetation cover in the Brazilian Atlantic Forest (Rezende et al. 2018), reflecting improvements compared to earlier estimates (Ribeiro et al. 2009). However, the effectiveness of such efforts may still not be optimal, as crucial flora and funga components might not receive adequate protection within designated zones, or their presence in PAs may be overlooked or undocumented at local or regional scales (e.g., Oliveira et al. 2017; Colli-Silva et al. 2019).

Ongoing efforts have the potential to contribute to the development of botanical monographs and regional floras inside Brazilian PAs. Botanical monographs are a crucial resource to support the continuous updating of lists of threatened species (Grace et al. 2021) and, subsequently, the formulation of conservation policies (Giam et al. 2010). In Brazil, many PAs have a residual conservation character, often encompassing historically less utilized lands (Oliveira et al. 2017; Vieira et al. 2019). As a result, a significant number of preserved specimen collections may have been gathered outside these PAs (Oliveira et al. 2017). This situation can lead to incomplete understanding of species biology, conservation status, and biogeography within and outside protected lands.

Among the groups potentially affected by this scenario is Rutaceae (Sapindales), the citrus family, known for its diverse growth forms and morphologies (Groppo et al. 2022). One of Rutaceae's primary centers of species richness and endemism lies within the Atlantic Forest (Colli-Silva and Pirani 2019). The state of Espírito Santo, in Southeastern Brazil, also stands out for its particularly high diversity of Rutaceae species (Colli-Silva and Pirani 2019, 2022). Unlike most Brazilian states, Espírito Santo is entirely within the Atlantic Forest domain, with over 6,500–7,700 plant and fungi species reported (Dutra et al. 2015; BFG [The Brazil Flora Group] 2021). This number is expected to increase as ongoing floristic projects continues to describe new species (Dutra et al. 2022). Espírito Santo also serves as a focal point for species richness and endemism among many taxonomic groups, driven by a complex interplay of factors from the region's natural history (Cabanne et al. 2008; Menini Neto et al. 2016; Garraffoni et al. 2017; Mercier et al. 2023).

In this study, we aimed to provide an overview of the Rutaceae within PAs of Espírito Santo. Our dataset was integrated with remote sensing data and the delimitations of Brazil's PAs. This approach allowed us not only to address species representativeness of Rutaceae in Espírito Santo but also to explore the representation of different taxa within and

outside the state's PAs. Our study is also tailored to serve as a resource in facilitating informed decisions by policymakers and taxonomists engaged in the study of the flora of Espírito Santo.

2 Material and methods

Occurrence data compilation and curation — Our occurrence dataset was obtained from the Global Biodiversity Information Facility platform (GBIF), which aggregates data from various local and regional repositories in Brazil, as well as collections from other international sources (Robertson et al. 2014). For the primary reference for native Rutaceae species reported in Espírito Santo, we relied on Brazil's Flora and Funga (BFG [The Brazil Flora Group] 2021; see also Colli-Silva and Pirani 2022). Our occurrence dataset focuses exclusively on preserved specimen collections, with an emphasis on collections identified at the species level. This dataset was previously compiled by Colli-Silva and Pirani (2019), who conducted a survey on the biogeographical patterns of Rutaceae and identified areas of endemism, utilizing an expert curated dataset of Rutaceae occurrence records in Brazil.

In cases where voucher labels lacked coordinates, we applied the georeferencing framework outlined by Magdalena et al. (2018). For conservation statuses, we consulted primarily the global IUCN (the International Union for Conservation of Nature) red list. We evaluated the number of "locations," as defined by IUCN latest guidelines (IUCN 2024), which refers to distinct geographic or ecological areas with a potential threat to all individuals of a specific taxon. Venn diagrams illustrating the conservation status, and threat levels were constructed in R Environment, using "ggVennDiagram" v. 1.2.2 package (Gao et al. 2021; R Core Team 2022).

Remote sensing data – Satellite images with a 30-m resolution were obtained from the INPE (Brazil's National Institute of Spatial Research) database, covering July 2019 to May 2021. Images with less than 10% cloud coverage were prioritized to ensure optimal terrain visualization. Considering Espírito Santo's area of 46,095 km², the region was divided into 185 km² quadrants for detailed analysis. Therefore, we obtained images from six quadrants from the INPE catalog: 215/73, 215/74, 216/72, 216/73, 216/74, and 216/75.

The downloading and gathering of remote sensing data were made for bands 2, 3, 4, and 5, corresponding to blue, green, red, and near-infrared reflectance values. Analyses were performed in QGIS (www.qgis.org), and NDVI (Normalized Difference Vegetation Index) values were extracted for all state boundaries. NDVI, a standard index for assessing vegetation health, density, and coverage, is calculated



as the difference between near-infrared and red reflectance values divided by their sum (Rouse et al. 1974). NDVI values typically range from -1 to +1, with negative values indicating water bodies, values near zero representing barren lands, and higher values indicating various types of vegetation from grasslands ($0 \le \text{NDVI} < 0.2$) to closed-canopy forests ($0.2 \le \text{NDVI} \le 1$) (following thresholds used in Freitas and Cruz 2003).

NDVI is a valuable metric for distinguishing land cover types, monitoring temporal vegetation changes, and assessing the impacts of land use (Freitas and Cruz 2003; Jensen 2007). Continuous NDVI values were cross-referenced with the Protected Areas network shapefile from The World Database on Protected Areas (UNEP-WCMC and IUCN 2024) in QGIS. We performed a Mann–Whitney U test in R (R Core Team 2022) to statistically compare average NDVI profiles of species occurrence records inside and outside PAs, under a significance level of 0.05.

3 Results

Occurrence dataset – We found a total of 62 species native to Brazil in 17 genera of Rutaceae having occurrence records within Espírito Santo state, although the current reports in Brazil's Flora and Funga is 56 species. *Balfourodendron riedelianum* (Engl.) Engl., *Conchocarpus odoratissimus* (Lindl.) Kallunki & Pirani, *Conchocarpus pentandrus* (A. St.-Hil.) Kallunki & Pirani, *Ertela trifolia* (L.) Kuntze, and *Zanthoxylum riedelianum* Engl. have confirmed occurrence records in Espírito Santo. We compiled 2,386 occurrence records with confirmed species-level identification from the initial dataset of 117 species and 3,720 unique observations originally compiled by Colli-Silva and Pirani (2019) (Table 1). A comprehensive list of all specimen records, following data cleaning and validation, is provided as Supplementary Material.

The identification phase of specimens was crucial for analyzing extinction risks and occurrence locations. Challenges from the GBIF-mobilized data included incomplete names, information inconsistencies, incorrect identifications (often made by non-expert taxonomists), and duplicate records with differing taxonomy. Likewise, most of the georeferencing issues found in our survey included vouchers with missing information on the label, and for those with coordinates already assigned, we identified a few cases with inverted coordinates or georeferencing errors, which we had to correct manually on a case-by-case basis.

Occurrence profile inside and outside PAs – The majority of occurrence records in our dataset were located outside the PAs network of Espírito Santo (1634 records, 68% of

the 2,386 reported and georeferenced occurrences) (histograms of Fig. 2). Occurrence records within PAs generally had higher levels of NDVI (Table 1; histograms of Fig. 2), and the distribution of the occurrence records had varied significantly when considering especially records that felt ouside PAs (see map on Fig. 2).

Regarding species representation, the vast majority (86%, 41 out of 62 species) had at least one preserved specimen occurrence within the PAs of the state (Table 1). Among the species assessed by IUCN for threat status, seven were classified as threatened with extinction (Fig. 2a). One species, *Spiranthera atlantica* Pirani, endemic to Espírito Santo, has been designated as "Data Deficient" due to limited occurrence records, yet it is found within PAs of Espírito Santo (Fig. 2a).

There are 21 species not found within any of the known PAs of Espírito Santo (Fig. 2b). Of these unrepresented species, six are endemic to the state and/or face threats of extinction, all belonging to the genus *Conchocarpus* J.C.Mikan (Fig. 2, Table 1): *Conchocarpus albiflorus* (Bruniera & Groppo) Bruniera & Groppo (also listed as "Endangered" in the IUCN red list), *C. bellus* Kallunki, *C. cauliflorus Pirani*, *C. furcatus* Kallunki (also listed as "Critically Endangered"), *C. macrocarpus* (Engl.) Kallunki & Pirani (also listed as "Endangered") and *C. minutiflorus* Groppo & Pirani (also listed as "Critically Endangered").

4 Discussion

Species representation within PAs – The residual character of PAs—In Brazil, PA delimitations have often prioritized the viability of designated areas over the diversity and biological aspects of the species present (Oliveira et al. 2017; Vieira et al. 2019). Oliveira et al. (2017) found that over half of the Brazilian species described to date and approximately 40% of evolutionary lineages lack protection from the infrastructure of Brazil's PAs. This issue may be reflected in the case of Rutaceae in Espírito Santo, where biogeographical aspects of species have not been fully considered in PA planning.

Another aspect to consider for conservation is phylogenetic diversity, which encompasses the evolutionary history of the lineages. Our results show that at least one species from each genus occurs within Espírito Santo's PAs. However, half of the species from *Conchocarpus*, *Ertela* Adans., *Esenbeckia* Kunth, and five out of nine *Zanthoxylum* L. species are not found in PAs in Espírito Santo. Notably absent species are *Metrodorea stipularis* Mart., with its unique characters, and *Pilocarpus pauciflorus* A.St.-Hil., which requires further studies to elucidate its evolutionary relationships (Pirani and Groppo 2020).



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Table 1 Occurrence, endemism, and threat status profile of Rutaceae species within and outside the protected areas (PAs) network in Espírito Santo state (ES), Southeastern Brazil

Angostura bracteata (Nees & Mart.) Kallunki Balfourodendron riedelianum (Engl.) Engl. Conchocarpus adenantherus (Rizzini) Kallunki & Pirani Conchocarpus abliftorus (Bruniera & Groppo) Bruniera & Groppo Conchocarpus bellus Kallunki Conchocarpus caulifforus Pirani Conchocarpus diadematus Pirani Conchocarpus diadematus Pirani Conchocarpus gelegans (A. StHil.) Kallunki & Pirani Conchocarpus furcatus Kallunki Conchocarpus heterophyllus (A. StHil.) Kallunki & Pirani Conchocarpus macrocarpus (Engl.) Kallunki & Pirani Conchocarpus macrocarpus (Engl.) Kallunki & Pirani Conchocarpus macrocarpus (Engl.) Kallunki & Pirani Conchocarpus macrocarpus (Rizzini) Kallunki & Pirani	1 Inside PAs 54 2 7 7 0 0 0	Outside PAs							status
All 107 107 107 11 11 11 12 23 6 11 11 11 11 11 11 11 11 11 11 11 11 1	Inside PAs 54 7 0	Outside PAs							(ICCN)
ni 177 17 17 18 & Groppo 19 9 17 17 23 6 6 11 11 11 11 10 11 11 11 11 11 11 11 11	45 2		All	Inside PAs	Outside PAs	p-value	Brazil	ES	
17 a & Groppo 19 9 17 17 17 18 19 19 10 irani 68 mi 60 mi 46 16	2 7 7 0	53	0.552 ± 0.334	0.836 ± 0.013	0.262 ± 0.242	*	Y	z	rc
ni 11 a & Groppo 19 9 17 23 6 11 11 10 irani 68 ni 60 17 17	7 0 0	15	0.430 ± 0.273	0.800 ± 0.000	0.381 ± 0.251	ı	z	z	EN
a & Groppo 19 9 17 23 6 6 11 11 11 10 hrani 68 nni 66	0 0	4	0.648 ± 0.284	0.822 ± 0.004	0.344 ± 0.274	*	Y	z	EN
9 17 23 6 6 11 11 11 10 60 mi 60 17 17 17 17 17 17 17 17 17 17 17 17 17	0	19	0.317 ± 0.181	ı	0.317 ± 0.181	1	Y	z	EN
17 23 6 1 11 10 irani 60 mi 46 166 17		6	0.668 ± 0.142	ı	0.668 ± 0.142	1	Y	Y	NE BE
23 6 1 11 10 irani 68 ni 60 17	0	17	0.332 ± 0.201	ı	0.332 ± 0.201	1	Y	Y	NE E
6 1 11 10 hrani 68 nni 60 17	5	18	0.282 ± 0.322	0.845 ± 0.022	0.125 ± 0.121	*	Y	z	NE BE
1 11 10 irani 68 uni 60 166 17	0	9	0.516 ± 0.309	ı	0.516 ± 0.309	1	Y	z	ГС
11 10 10 mi 68 16 16	0	1	I	ı	ı	1	Y	z	NE
10 irani 68 uni 60 166 17	0	111	0.359 ± 0.056	ı	0.359 ± 0.056	1	Y	z	NE
hrani 68 nni 60 166 17	0	10	0.446 ± 0.333	ı	0.446 ± 0.333	1	Y	Y	CR
mi 60 46 166 17	19	49	0.420 ± 0.272	0.757 ± 0.170	0.289 ± 0.172	*	Y	z	ГС
46 166 17	29	31	0.569 ± 0.266	0.707 ± 0.233	0.439 ± 0.231	*	z	z	ГС
166	0	46	0.299 ± 0.264	ı	0.299 ± 0.264	1	Y	z	EN
17	33	133	0.570 ± 0.270	0.828 ± 0.021	0.506 ± 0.265	*	Y	z	NE
_	3	14	0.367 ± 0.302	0.837 ± 0.002	0.267 ± 0.225	*	Y	Y	NE
	0	1	ı	ı	I	ı	Y	z	NE
Conchocarpus minutiflorus Groppo & Pirani	0	18	0.433 ± 0.159	I	0.433 ± 0.159	ı	Y	Y	CR
Conchocarpus obovatus (Nees & Mart.) Kallunki & Pirani 22 9	6	13	0.689 ± 0.213	0.813 ± 0.010	0.603 ± 0.245	*	Y	z	NE
Conchocarpus odoratissimus (Lindl.) Kallunki & Pirani	0	3	0.166 ± 0.063	ı	0.166 ± 0.063	ı	Y	z	NE
Conchocarpus pentandrus (A. StHil.) Kallunki & Pirani	2	1	0.601 ± 0.423	0.845 ± 0.035	I	I	z	z	NE
Conchocarpus punctatus Kallunki	4	0	0.825 ± 0.026	0.825 ± 0.026	I	1	Υ	z	NE
Conchocarpus ruber (A.St.Hil.) Bruniera & Groppo	40	220	0.540 ± 0.267	0.835 ± 0.018	0.486 ± 0.255	*	z	z	NE
Conchocarpus silvestris (Nees & Mart.) Kallunki & Pirani 18	1	17	0.385 ± 0.197	I	0.359 ± 0.170	ı	¥	z	NE
Dictyoloma vandellianum A. Juss	41	55	0.651 ± 0.227	0.764 ± 0.121	0.567 ± 0.251	*	z	z	ГС
Dryades cyrtantha (Kallunki) Groppo & Kallunki	22	6	0.771 ± 0.164	0.839 ± 0.018	0.605 ± 0.238	*	¥	z	NE
Dryades insignis (Pirani) Groppo & Pirani	14	16	0.458 ± 0.153	0.402 ± 0.048	0.507 ± 0.195	*	¥	z	NE
Ertela bahiensis (Engl.) Kuntze	2	1	0.587 ± 0.357	0.793 ± 0.000	I	ı	Y	z	NE
Ertela trifolia (L.) Kuntze 5 (0	5	0.644 ± 0.323	I	0.644 ± 0.323	I	z	z	NE
Erythrochiton brasiliensis Nees & Mart.	12	94	0.596 ± 0.228	0.835 ± 0.034	0.565 ± 0.224	*	z	z	ГС
Esenbeckia febrifuga (A.StHil.) A. Juss. ex Mart.	0	18	0.657 ± 0.231	I	0.657 ± 0.231	ı	z	z	NE
Esenbeckia grandiflora Mart. 59	59	93	0.564 ± 0.254	0.706 ± 0.188	0.474 ± 0.250	*	z	z	ГС
Esenbeckia leiocarpa Engl.	3	19	0.617 ± 0.280	0.857 ± 0.004	0.579 ± 0.284	*	Y	Z	ΛΩ



Table 1 (continued)

Species	Occur	Occurrence records in ES	in ES	NDVI (mean±s.d.)	s.d.)			Endemism	ws.	Threat status (TUCN)
	All	Inside PAs	Outside PAs	All	Inside PAs	Outside PAs	p-value	Brazil	ES	
Esenbeckia pilocarpoides Kunth	3	0	3	0.827 ± 0.000	ı	0.827 ± 0.000	1	z	z	LC
Galipea carinata Pirani	30	17	13	0.576 ± 0.169	0.534 ± 0.074	0.631 ± 0.237	*	Y	Y	NE
Galipea jasminiflora (A.StHil.) Engl.	51	24	27	0.653 ± 0.265	0.825 ± 0.027	0.500 ± 0.287	*	Y	Z	ГС
Galipea laxiflora Engl.	45	24	21	0.652 ± 0.255	0.817 ± 0.038	0.464 ± 0.268	*	Y	z	ГС
Hortia brasiliana Vand. ex DC.	26	14	12	0.730 ± 0.215	0.813 ± 0.047	0.634 ± 0.289	*	z	z	ГС
Metrodorea maracasana Kaastra	4	3	1	0.654 ± 0.343	0.826 ± 0.000	ı	ı	Y	z	NE
Metrodorea nigra A.StHil.	36	24	12	0.693 ± 0.259	0.828 ± 0.006	0.425 ± 0.310	*	Y	Z	ГС
Metrodorea stipularis Mart.	2	0	2	0.188 ± 0.000	I	0.188 ± 0.000	ı	Y	Z	ГС
Neoraputia alba (Nees & Mart.) Emmerich ex Kallunki	143	57	98	0.619 ± 0.279	0.837 ± 0.017	0.474 ± 0.276	*	Y	z	ГС
Neoraputia magnifica (Engl.) Emmerich ex Kallunki	43	31	12	0.688 ± 0.233	0.794 ± 0.102	0.416 ± 0.255	*	Y	Z	ГС
Pilocarpus giganteus Engl.	8	4	4	0.808 ± 0.020	0.826 ± 0.000	0.790 ± 0.007	*	Y	Z	NE
Pilocarpus grandiflorus Engl.	20	8	12	0.551 ± 0.285	0.821 ± 0.013	0.370 ± 0.227	*	Y	Z	NT
Pilocarpus grandiflorus Engl. var. grandiflorus	5	_	4	0.412 ± 0.312	I	0.309 ± 0.242	ı	Y	z	NE
Pilocarpus pauciflorus A.StHil.	7	0	7	0.424 ± 0.299	I	0.424 ± 0.299	ı	Y	Z	LC
Pilocarpus riedelianus Engl.	52	15	37	0.583 ± 0.258	0.813 ± 0.065	0.489 ± 0.248	*	Y	z	LC
Pilocarpus spicatus A.StHil.	77	18	59	0.660 ± 0.220	0.722 ± 0.187	0.641 ± 0.228	us	Y	z	LC
Rauia nodosa (Engl.) Kallunki	91	43	48	0.541 ± 0.250	0.663 ± 0.226	0.431 ± 0.218	*	Y	z	LC
Rauia resinosa Nees & Mart.	72	20	52	0.545 ± 0.302	0.823 ± 0.019	0.438 ± 0.291	*	z	Z	ГС
Ravenia infelix Vell.	87	37	50	0.640 ± 0.253	0.805 ± 0.088	0.518 ± 0.266	*	Y	z	NT
Spiranthera atlantica Pirani	10	6	1	0.830 ± 0.014	0.826 ± 0.000	I	ı	Y	×	DD
Zanthoxylum acuminatum (Sw.) Sw.	28	14	14	0.629 ± 0.291	0.823 ± 0.011	0.435 ± 0.308	*	Z	z	ГС
Zanthoxylum caribaeum Lam.	11	0	111	0.254 ± 0.194	ı	0.254 ± 0.194	ı	Z	z	ГС
Zanthoxylum fagara (L.) Sarg.	8	0	∞	0.352 ± 0.167	I	0.352 ± 0.167	ı	z	z	TC
Zanthoxylum monogynum A.StHil.	69	12	57	0.486 ± 0.233	0.682 ± 0.210	0.445 ± 0.217	*	Z	z	TC
Zanthoxylum nemorale Mart.	2	0	2	0.720 ± 0.094	ı	0.720 ± 0.094	ı	¥	z	ГС
Zanthoxylum petiolare A.StHil. & Tul.	1	0	1	I	I	I	ı	z	z	TC
Zanthoxylum rhoifolium Lam.	63	10	53	0.612 ± 0.254	0.798 ± 0.057	0.577 ± 0.262	*	z	z	TC
Zanthoxylum riedelianum Engl.	2	0	2	0.778 ± 0.000	I	0.778 ± 0.000	ı	Z	z	ГС
Zanthoxylum tingoassuiba A.StHil.	10	9	4	0.603 ± 0.237	0.684 ± 0.220	0.482 ± 0.235	ns	¥	z	ГС
All species	2386	752	1634	0.566 ± 0.272	0.776 ± 0.139	0.470 ± 0.264	*	I	ı	ı

The *p*-values indicate the results of Mann–Whitney U tests (see Material and Methods), assessing the statistical differences in NDVI values between the two categories (inside and outside PAs). Statistical significance is denoted as * for differences at the 0.05 significance level and "ns" for non-significant results. Species endemism is indicated for Brazil and Espfrito Santo (Y = yes; N = no). Extinction risk is categorized based on IUCN red list categories: LC = Least Concern; NT = Near Threatened; EN = Endangered; VU = Vulnerable; CR = Critically Endangered; DD = Data Deficient. A full list of all occurrence records can be found in Supplementary Material (Appendix S1) The table differentiates occurrence records based on their presence inside and outside PAs and presents the mean NDVI values calculated for each species (mean ± standard deviation, s.d.).



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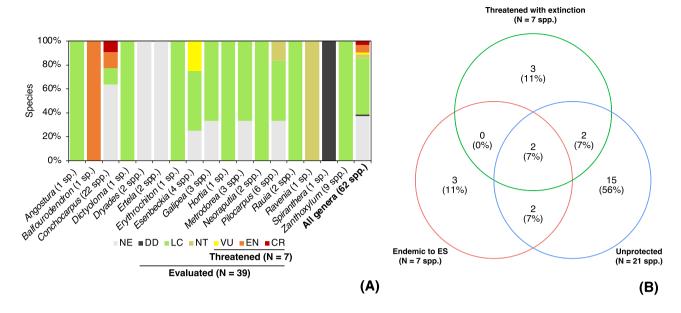


Fig. 1 Overview of (A) threat status distribution across Rutaceae genera in Espírito Santo state, and (B) species count by category, including those threatened with extinction and those protected or unprotected (i.e., having at least one specimen record documented within any protected area of Espírito Santo state). The figure also includes data on species endemism to Espírito Santo

Most authors recommend expanding PA networks to include areas of high taxonomic and phylogenetic uniqueness (Oliveira et al. 2017; Saraiva et al. 2018). This includes maintaining the rate of field expeditions, improving funding, and prioritizing sampling in PAs where little is known about forest structure and biodiversity (Colli-Silva et al. 2016; Oliveira et al. 2017; Saraiva et al. 2018). The low representation of species in PAs may also result from limited collection efforts, especially given the challenging terrain of many PAs in the Atlantic Forest, often characterized by steep slopes that hinder access and fieldwork, as discussed by Vieira et al. (2019).

In terms of species representation within and outside PAs, Colli-Silva et al. (2019) reported that, for seed plants from the state of São Paulo, out of 8,521 recorded species, 48% occur in PAs, 361 are endemic to the state, and 676 are threatened with extinction. However, 65% of these threatened species are unprotected, with 51 of them endemic to São Paulo, primarily native to the Atlantic Forest. In Espírito Santo, we showed that 21 out of 62 Rutaceae species (33% of the total) have no records in protected areas. For Rutaceae, the situation would be supposedly slightly worse in São Paulo, where Colli-Silva et al. (2019) documented 43 species, with 16 species (37%) lacking records within the state's PAs. Unlike São Paulo, where the two threatened species are not endemic to the state, Espírito Santo has six Conchocarpus species that are both endangered and endemic (see below), presenting a more critical situation. However, unlike São Paulo, Espírito Santo has six Conchocarpus species that are both endangered and endemic, presenting

a more critical situation. After reviewing their conservation status, we actually confirmed that no Rutaceae species in São Paulo are currently listed as threatened with extinction, although four species are classified as Near Threatened: *Balfourodendron riedelianum* (Engl.) Engl., *Esenbeckia hieronymi* Engl., *Hortia brasiliana* Vand. ex DC., and *Pilocarpus giganteus* Engl.

The unprotected Conchocarpus—All six threatened Conchocarpus species are situated within degraded areas of dense ombrophyllous forest or semi-deciduous seasonal forests, as sciophilous plants inhabiting the understory. Among these species, only populations of C. macrocarpus have been observed in both lowland and submontane formations; the remaining species are distributed across boundaries between these two types of formations (Kallunki and Pirani 1998; Pirani and Groppo 2020). These Conchocarpus are found at elevations between 55 and 313 m, on slopes (C. albiflorus and C. furcatus), particularly slopes of granite inselbergs (C. bellus and C. macrocarpus), plateaus (C. cauliflorus, C. macrocarpus), and lowlands (C. minutiflorus).

While the population of *C. albiflorus* is found within a strict protected area in Rio de Janeiro, it inhabits a severely fragmented remnant of seasonal semi-deciduous forest in Espírito Santo (Bruniera et al. 2015, 2021; Pirani and Groppo 2020; Fernandez et al. 2021). This species is classified as Endangered (EN) due to its limited and fragmented distribution and ongoing habitat decline (Fernandez et al. 2021). Compared to *C. ruber* (A.St.-Hil.) Bruniera & Groppo, its closest relative (Bruniera et al. 2015, 2021),



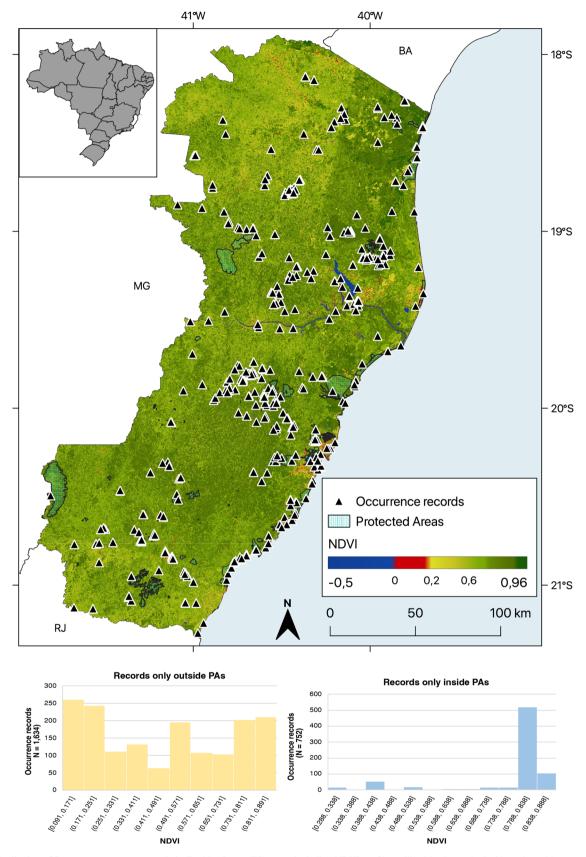


Fig. 2 Distribution of Rutaceae occurrence records in Espírito Santo (ES) state, including NDVI profiles utilized in the study, inside and outside protected areas network. The figure includes histograms displaying the NDVI profile distribution for all occurrence records, highlighting the distinction between records inside and outside the state's protected areas network. Adjacent regions (Brazilian states) represented: Minas Gerais (MG), Bahia (BA), and Rio de Janeiro (RJ)



C. albiflorus occupies less healthy forest remnants and has smaller population sizes.

Among the other species endemic to Espírito Santo, *C. bellus* is critically endangered (CR) due to agricultural activities, forestry, and livestock farming, threatening its sole population from Espírito Santo's center-north (Pirani et al. 2013). Currently, *C. bellus* is not included in any protected area of the state, though the nearest protected area close to one occurrence record is the Reserva Biológica de Sooretama and Reserva Natural Vale.

Conchocarpus cauliflorus is closely related to C. macrocarpus and C. obovatus, sharing inflorescences with sessile cymules and one to three flowers per branch. Despite their phylogenetic proximity, only C. obovatus is found in protected areas and exhibits higher NDVI scores. No threats to C. obovatus were identified by us, while C. cauliflorus and C. macrocarpus face pressures from agriculture, livestock, and silviculture (Pirani et al. 2013; Moraes et al. 2020).

Finally, two other species endemic to Espírito Santo are *C. furcatus* and *C. minutiflorus* (Pirani and Groppo 2020). *Conchocarpus furcatus* is distinctive due to its partial inflorescences (dichasial proximally and monochasial distally) (Kallunki and Pirani 1998), while *C. minutiflorus* has perennial, pauciflorous inflorescences (Pirani et al. 2011). Both are considered critically endangered (CR) due to their restricted distribution in fragmented forests (Fernandez et al. 2020a, b).

Insights from remote sensing data — Our findings highlight the value of remote sensing in monitoring conservation, guiding new collections, and identifying regions for potential protection. Species found exclusively within PAs had the highest NDVI values, emphasizing the role of PAs in preserving healthy habitats. This was statistically significant for genera like *Conchocarpus*, *Metrodorea*, *Pilocarpus*, and *Zanthoxylum*. However, *Esenbeckia* was an exception, with no significant difference between protected and unprotected occurrences, and higher NDVI values also seen for unprotected species. This discrepancy may be due to incomplete data for certain PAs, like Parque Natural Municipal Morro da Pescaria.

Remote sensing has become a valuable tool for assessing vegetation quality and preservation (Holm 2003; Barbosa et al. 2006). In our study, it revealed significant heterogeneity in vegetation cover across Espírito Santo, highlighting its importance for monitoring plant populations, especially those at risk of extinction. The integration of NDVI values allowed us to assess forest health and density associated with Rutaceae species. Similar approaches have been used to assess plant vulnerability (Pesaresi et al. 2020; Matas-Granados et al. 2022) and identify conservation hotspots, habitat conditions, or species diversity (Nagendra et al. 2013; Dubinin et al. 2018; Silveira et al. 2021). Additionally, the

integration of remote sensing enhances our understanding of vegetation health and distribution, particularly for species outside PAs. It informs conservation planning by prioritizing areas with high biodiversity and robust ecosystems. By identifying regions with healthy vegetation and critical Rutaceae populations, remote sensing is a powerful tool for guiding conservation efforts and improving biodiversity preservation strategies in Espírito Santo.

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Data availability All data supporting the findings of this study are included in the article and its supplementary materials. Additionally, remote sensing data used in the study are available for download from public databases, as detailed in the Methods section.

Declarations

Conflict of interest The authors declare no conflict of interest.

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